

## New Approaches in the Treatment of Osteoarthritis

Belén Cuervo, Mónica Rubio, Ramón Cugat, Montserrat García-Balletbó, Elena Damiá, Joaquín J Sopena, Jose M<sup>a</sup> Carrillo

Belén Cuervo, Mónica Rubio, Ramón Cugat, Montserrat García-Balletbó, Joaquín J Sopena, Jose M<sup>a</sup> Carrillo, García Cugat Chair of Surgery and Regenerative Medicine of Universidad CEU Cardenal Herrera. Valencia, Spain

Belén Cuervo, Mónica Rubio, Ramón Cugat, Montserrat García-Balletbó, Elena Damiá, Joaquín J Sopena, Jose M<sup>a</sup> Carrillo, García Cugat Foundation. Barcelona, Spain

Belén Cuervo, Mónica Rubio, Elena Damiá, Joaquín J Sopena, Jose M<sup>a</sup> Carrillo, Medicine and Veterinarian Surgery Department, Universidad CEU Cardenal Herrera, Valencia, Spain

Mónica Rubio, Joaquín J Sopena, Jose M<sup>a</sup> Carrillo, Biomedical Science Institut, Universidad CEU Cardenal Herrera, Valencia, Spain

Cugat R, García-Balletbó M, GC Arthroscopy, Quirón Hospital, Barcelona, Spain

Elena Damiá, Loriguilla Veterinary, Valencia, Spain

Correspondence to: Belén Cuervo, García Cugat Chair of Surgery and Regenerative Medicine of Universidad CEU Cardenal Herrera. Valencia, Spain

Email: [seijastraumatologia@gmail.com](mailto:seijastraumatologia@gmail.com)

Telephone: +34932172252

Fax: +34932381634

Received: September 13, 2014

Revised: October 11, 2014

Accepted: October 15, 2014

Published online: February 23, 2015

### ABSTRACT

Degenerative joint disease, also known as osteoarthritis, is currently one of the most prevalent and morbid diseases due to an increase in both life expectancy and obesity. Moreover, due to its severity and irreversibility, osteoarthritis causes a severe social and economic impact around the world due to the loss in quality of life and productivity. Osteoarthritis is known to be a multifactorial disease, where several risk factors have been already described, leading to

temporary and progressive structural changes within joint tissues, especially in joint cartilage, subchondral bone, synovial membrane and synovial liquid, causing the main clinical symptoms such as chronic pain, joint instability, stiffness and joint space narrowing. The use of advanced therapies for treating this disease is guided to a regenerative medicine aimed to encourage the regenerative or restorative methods, being a new alternative to more aggressive treatments. Within these treatments we can find platelet-rich plasma (PRP) and mesenchymal stem cells (MSC). PRP is an autologous novel therapeutic tool that has risen strongly in the past few years and has several applications within the medical field. It is defined as a biological product derived from the own patient's blood, obtaining a plasma fraction after a centrifugation process with a platelet concentration greater than the existing in the circulatory system. Many of the factors contained in the interior of PRP have specific activity of neoproliferation and cartilage regeneration. There are increasing studies regarding MSC and osteoarthritis treatment, especially in relation to MSC derived from adipose tissue. These cells have shown its capacity to differentiate into chondrocytes, osteocytes and adipocytes, as well as their advantage that they can be obtained in bigger quantities with little morbidity to the donor site and reduced discomfort to the patient. It has been confirmed that autologous MSC have great affinity for the injured tissue of the joint, being able of localizing and participating in repair of structures of the injured joint.

© 2015 ACT. All rights reserved.

**Key words:** Osteoarthritis; PRGF; Mesenchymal cells

Cuervo B, Rubio M, Cugat R, García-Balletbó M, Damiá E, Sopena JJ, Carrillo JM. New Approaches in the Treatment of Osteoarthritis. *International Journal of Orthopaedics* 2015; 2(1): 210-218 Available from: URL: <http://www.ghrnet.org/index.php/ijo/article/view/1054>

## INTRODUCTION

Osteoarthritis (OA) is a very common degenerative disease affecting the articular cartilage in both human<sup>[1]</sup> and veterinary medicine<sup>[2,3]</sup>. It is a disabling disease characterized by progressive damage to the articular cartilage with involvement of the subchondral bone, osteophyte formation, thickening of the joint capsule and synovitis<sup>[4]</sup>, causing discomfort and pain in the affected joint, in many cases resulting in the implantation of a prosthesis as a final method of restoring function and decreasing pain<sup>[5]</sup>.

Joint cartilage is a highly specialized, hypocellular, avascular, alymphatic, not innervated, dense connective tissue. It contains a low percentage of cells in its composition (5%), and is embedded in an extracellular matrix rich in collagen, proteoglycans, other proteins and water<sup>[6]</sup>. As one ages, the density and capacity of dividing cells or chondrocytes present in articular cartilage decreases, limiting their capacity for self-renewal<sup>[7]</sup>. There comes a time when this division is weak, in such a way that aging contributes to the progressive loss of articular surface resulting in the process of cartilage degeneration with matrix loss, fibrillation and cracking, which can result in a complete loss of thickness of the joint surface<sup>[8,9]</sup>. Within the affected population, risk factors that predominate are age, being female, obesity and previous trauma<sup>[4]</sup>, and other causes that can trigger the disease, like improper loading, biochemical changes and genetic background<sup>[10-15]</sup>.

In dogs, this disease is a common disorder that affects dogs of all ages with a high incidence that affects 20% of the population of adult dogs and 80% of geriatric animals, and is the most common cause of lameness in dogs older than one year<sup>[16]</sup>. However, in the case of the feline species it is an underdiagnosed disease because in most cases cats show no lameness, but gradual changes in behavior and lifestyle, the main risk factor in this species is age<sup>[17]</sup>. In both species the joints

most affected by this disease are the elbow, hip and knee<sup>[3]</sup>.

An important factor to consider in the assessment and diagnosis of this condition is the lack of correlation in most cases between the clinical signs of the disease, and radiological signs because sometimes there is a degree of lameness in patients and very high pain which are virtually absent radiological signs. Conversely, patients with severe radiological signs showing no symptoms<sup>[4]</sup>. For this reason, treatment should be aimed at alleviating pain and inflammation while trying to stop the disease from advancing, thus increasing patients quality of life<sup>[1]</sup> (Figure 1).

Among the therapeutic modalities used to treat osteoarthritis there are several surgical and non-surgical treatments. Surgical treatments are used especially in the correction of the primary causes of osteoarthritis<sup>[18]</sup>. Among non-surgical treatments there are non-pharmacological measures such as weight reduction, therapeutic foods, low impact exercise programs and physiotherapy, and pharmacologic measures such as anti-inflammatory drugs, analgesics, corticosteroids, Disease-modifying antirheumatic drugs (DMARDs)<sup>[19-22]</sup> and the use of nutraceuticals and glycosaminoglycans<sup>[23]</sup>.

There are other therapies that are not in any of these categories such as the use of intra-articular botulinum toxin<sup>[24,25]</sup>, or autologous chondrocyte implantation<sup>[26]</sup>.

Today the term Advanced Therapies (AT) refers to these new therapies focusing on regenerative medicine that aims to promote regenerative or reparative phenomena on the degenerative. Among these therapies, are the so-called platelet-rich plasma (PRP) and mesenchymal cells (MSC)

## PLATELET RICH PLASMA

Platelet Rich Plasma (PRP) is defined as an autologous biological product derived from the patient's blood, and in which after a centrifugation process a plasma fraction is obtained with a platelet concentration higher than that in circulating blood<sup>[27]</sup>. The platelets contained in the plasma fraction can be artificially activated to release the contents contained within their granules. Among the substances contained within platelet granules there are various factors<sup>[28,29]</sup>.

Growth factors are a group of substances of polypeptide, soluble and diffusible nature, that are capable of regulating the growth, differentiation and phenotype of numerous cell types. They are synthesized by a wide variety of cell types, and can exert their effect on local cell metabolism or systemic circulation. In circulation, the growth factors can be found free from proteins or protein-bound, or may be found within the  $\alpha$  granules of platelets, which are released after the process of platelet activation<sup>[30,31]</sup>. Locally, growth factors can act in an autocrine (on the same cells that produce them) and/or paracrine (on other cell types located nearby) manner, regulating cellular functions<sup>[30,31]</sup>.

These growth factors provide the initial signals for the activation of the cells in the surrounding tissue. In response to the signals, local cells and infiltrated cells initiate changes in cell proliferation, differentiation and synthesis of proteins with diverse biological functions. Taken together, all these phenomena, define the process known as cell activation<sup>[32]</sup>. When these chemicals are released from platelets, multiplication and development of vascular endothelial cells, smooth muscle cells and fibroblasts becomes possible. Additionally exerting multiple effects on the phenomena of cellular remodeling<sup>[33]</sup> allowing reciprocal interaction between leukocytes and endothelial cells in order to modulate the inflammatory reaction and healing processes of tissue regeneration<sup>[34]</sup>.

Growth factors were first classified by Canalis in 1988<sup>[35]</sup>



**Figure 1** Ventrodorsal radiograph of the hip of a German Shepherd 33kg with bilateral degenerative joint disease.

describing transforming growth factor (TGF- $\beta$ ), bone-derived growth factor (BDGF), insulin-like growth factor I (IGF-I) and platelet-derived growth factor (PDGF) that are synthesized by skeletal cells. Vascular endothelial growth factor (VEGF) and epidermal growth factor (EGF), fibroblast growth factor, acidic and basic (aFGF & bFGF) have been isolated from bone matrix. Similarly, IGF-I and bFGF are synthesized from cartilage tissues, such as type I interleukins (IL-1). Tumor necrosis factor (TNF- $\alpha$ ), macrophage derived growth factors and PDGF are synthesized by blood cells. Subsequent studies have shown that these factors and many others are synthesized by many different cell types<sup>[30]</sup>.

A large number of these factors have a specific activity on neoproliferation, on cartilage regeneration and in particular also an antiapoptotic effect on chondroblasts. This has made the use of autologous growth factors in orthopedics a field of rapid growth and could have a crucial role in the treatment of osteoarthritis due to its influence on chemotaxis, differentiation, proliferation and cartilage synthetic activity and bone cells, thereby regulating physiological remodeling and cartilage repair<sup>[36]</sup>.

Some of the most important factors that act to help alleviate cartilage degradation in this disease are<sup>[36]</sup>: (1) PDGF regulates secretion and collagen synthesis; (2) EGF stimulates cell proliferation, endothelial chemotaxis and angiogenesis; (3) VEGF increases vascular permeability and angiogenesis; (4) TGF- $\beta$  stimulates the proliferation of undifferentiated mesenchymal cells, stimulates endothelial cell chemotaxis and angiogenesis; (5) bFGF promotes the growth and differentiation of chondrocytes and osteoblasts, stimulates mitogenesis of mesenchymal cells, chondrocytes and osteoblasts.

As discussed above, most of the growth factors are released upon platelet activation, which is why clinically it has become known as platelet rich plasma (PRP). The autologous nature of PRP, ease of implementation and relatively low cost, are some of the qualities that have popularized its increasing use in various fields of medicine and research<sup>[37]</sup>.

Platelet rich plasma preparations were first developed in the early 90s, and its first applications in cardiac surgery<sup>[38]</sup>. It was in the late 90's when it was introduced in oral surgery<sup>[39]</sup>. From then on it began to be used in various fields, such as general surgery, plastic surgery, neurosurgery, vascular surgery, ophthalmology, maxillofacial surgery and orthopedics, among others<sup>[40,41]</sup>.

In recent years there has been huge controversy regarding the nomenclature and definition of PRP<sup>[42,43]</sup>. According to Anitua and colleagues (2008), the term "Platelet Rich Plasma" is a vague and imprecise term. Within this term there a large number of autologous blood preparations that differ with regards to processing protocols and preparation, and therefore differ in quantitative and qualitative characteristics. Hence the importance of accurately and clearly defining the conditions of preparation and processing of PRP (platelet concentration, type of anticoagulant used, number and speed of centrifugation/same or different type(s) of platelet activator and of course the presence or absence of leukocytes)<sup>[43]</sup>.

Among the various plasma preparations one will find Plasma Rich in Growth Factors or PRGF<sup>®</sup>, this is the most advanced autologous PRP system, marketed by the Biotechnology Institute, BTI. It has CE certification for European health authorities, and is approved in Europe for the production of plasma growth factors and their application in various medical specialties. In the USA it is marketed under the name PRGF-Endoret<sup>®</sup>, and is a pioneer in the development of specific protocols for tissue regeneration as well as being the first 100% autologous technique on the market. Its advantages include<sup>[44]</sup> (Figure 2): (1) Biocompatible, versatile and safe; (2) Controlled



**Figure 2** Intraarticular infiltration of PRGF in hip.

activation and use. The activation process of platelets is promoted by the addition of calcium chloride, while causing the release of large amounts of growth factors and fibrin polymerization; (3) Simple protocol: single centrifugation, obtaining red cells at the bottom of the tube, just above are the white cells and then two fractions of plasma, the upper 60% corresponds to the fraction poor in growth factors, and below this the remaining 40% corresponds to plasma rich in growth factors; (4) Fast Protocol: 8 min centrifugation at 460g and 20 minutes to prepare; (5) Does not contain leukocytes, thus preventing pro-inflammatory activity; (6) It has bacteriostatic properties; (7) Great therapeutic potential, no side effects, significantly reduces recovery time of fractures, muscle injuries, tendons and surgery.

## PRGF IN OSTEOARTHRITIS

Effective treatment in degenerative joint disease remains a daunting clinical challenge today despite the advances in medicine. The aims when treating a pathology are to control pain, improve function and slow down the progression of the disease<sup>[45]</sup>.

The application of PRGF in joints with osteoarthritis is destined to trigger, promote and imitate the physiological process of regeneration and repair that are initiated spontaneously in tissues after injury. This process is based mainly on the function carried by the protein intervening in cellular communication, transmitting the information to interact with cell membrane receptors, leading to directed cell migration, proliferation and differentiation including, increasing cell activity that would occur physiologically<sup>[46-48]</sup>.

As it is autologous, bioresorbable, biocompatible and free of leukocytes and red blood cells, it is one of the best elements used in tissue engineering<sup>[49]</sup>. PRGF intraarticular infiltration is used in patients suffering from degenerative joint disease, and also brings biologically balanced growth factors, cytokines, and morphogens also contained in platelets and fibrinogen and other plasma proteins to the area<sup>[50,51]</sup>. Once activated PRGF is injected into the damaged joint, resulting in a gradual and uniform distribution in the affected area and becomes a viscous, malleable matrix. Furthermore, PRGF also



provides a "biological scaffold" or three-dimensional network in the joint composed of fibrin that is derived from the polymerization of fibrinogen. This network contains binding sites for cell adhesion, as well as proteins that form the microenvironment leading to different adhesion molecules and cells that help biological cartilage repair<sup>[52]</sup>.

There are several studies supporting the efficacy of this treatment. Examples of studies in human medicine are those of Wang *et al.*, which found a significant improvement in function and quality of life of patients with osteoarthritis of the knee at 6 months post treatment<sup>[53]</sup>, or Sanchez and colleagues, who compared the efficacy of intraarticular infiltration of PRGF compared with hyaluronic acid, obtaining more favorable results in the group treated with PRGF 5 weeks after treatment<sup>[54]</sup>. In veterinary medicine, there have been more studies with PRP, but there are some with PRGF. Satisfactory results have also been obtained in other works, such as those by Kwon *et al.* that showed macroscopic and histological improvements in rabbits with degenerative joint disease treated with PRP<sup>[55]</sup>, as well as Serra and colleagues who found that damaged tissue treated with autologous PRP showed a positive trend toward repairing over time<sup>[56]</sup>. Another study by Milano *et al.* also supports the efficacy of this treatment in sheep models suffering from the disease<sup>[57]</sup>. In dogs, studies have also been conducted using PRGF, evaluating the efficacy of intra-articular injection in animals with arthritic pathology in elbow, hip and knee, resulting in an improvement in of the disease<sup>[58,59]</sup>, and Silva and colleagues have shown that after ACL surgery there weren't any osteoarthritic changes in patients treated with platelet-rich plasma<sup>[60]</sup>.

For all of the aforementioned, PRGF plays a crucial role in the treatment of OA by promoting the repair and remodeling of injured tissue and preventing cartilage degradation and atrophy of the peri articular structures<sup>[49]</sup>.

## MESENCHYMAL CELLS

In recent years stem cells have generated enormous expectations and have become a great hope for the development of new cell therapies in the context of regenerative medicine<sup>[7,61]</sup>.

Stem cells are undifferentiated cells that have the ability to divide indefinitely without losing their properties and eventually produce specialized cells<sup>[7]</sup>.

They can be classified according to their capacity for differentiation or by their origin. According to differentiation capacity they are divided into<sup>[62]</sup>: (1) Totipotent: Ability to divide and produce all of the differentiated cells in an organism, including themselves; (2) Pluripotent: Give rise to any of the three germ cell layers ectoderm, endoderm and mesoderm and to other pluripotent cells, but they cannot give rise to less differentiated totipotent cells; (3) Multipotent: Give rise to different cell types of the same layer or embryonic lineage. (For example, a mesenchymal stem cell from bone marrow has a mesodermal nature and will give rise to cells of that layer such as myocytes, adipocytes or osteocytes, etc.); (4) Unipotent: They can only form a particular cell type.

According to their origin in<sup>[62]</sup>: (1) Embryonic: Are pluripotent cells and obtained from the early stages of embryonic development, from the inner cell mass of the blastocyst, embryo pre-implantation stage, 5-6 days after fertilization in which there are approximately 150 to 200 cells<sup>[63,64]</sup>. (2) Germline cell: The germ cells are pluripotent embryonic stem cells derived from embryonic gonadal regions, particularly in a specific area of the embryo called the gonadal ridge that gives rise to ovules and sperm. They have a capacity similar to that of embryonic stem cell differentiation, but their isolation is

more difficult<sup>[65]</sup>. (3) Fetal Stem Cells are derived from fetal tissues and organs such as blood, liver, and lung and possess characteristics similar to their counterparts in adult tissues. Have more expansion and differentiation capacity, although there are many doubts about the origin of these cells. Their origin could be embryonic cells which appear distributed throughout fetal tissues such as stem cell reservoirs or they could come from new waves of progenitors unrelated with embryonic cells<sup>[66]</sup>. (4) Somatic or adult cells are collected from any tissue in the adult organism resulting from the three germ layers and are capable of repair or regeneration of tissues such as placenta and umbilical cord<sup>[67,68]</sup>. Initially, it was believed that these cells were multipotent, i.e. could only differentiate in cell types derived from the same embryonic lineage they came from. However, in recent years there have been several studies that have provided surprising results, which suggest that the potential of some types of ASC is higher than expected, as they have shown under certain conditions the capacity to differentiate into different cell lineages<sup>[69-71]</sup>. **The can differentiate into:** (a) Hematopoietic stem cells, which give rise to all hematopoietic lineage cells, both the lymphoid (lymphocytes and natural killer cells) and myeloid (erythrocytes, platelets, monocytes, etc.); (b) Mesenchymal stem cells, which give rise to cells of different non-hematopoietic tissues such as chondrocytes, osteocytes, adipocytes, etc. (5) Induced pluripotent stem cells (iPS) are cells with embryonic characteristics that have been obtained by reprogramming adult cells. From the generation of such cells, the stem cell field has grown exponentially. These cells have become the main tools in the advancement of personalized medicine, as they emerged in order to avoid the ethical and technical issues in the generation of embryonic stem cells<sup>[72]</sup>.

Regarding stem cells of adult origin, within which are mesenchymal stem cells that are a very good option for various diseases as they are present in a large number of organs and postnatal connective tissue, and are not subject to the same restrictions as embryonic stem cells<sup>[73]</sup>. These cells possess a number of characteristics that define them<sup>[62,74]</sup>: (1) Capacity for self-renewal, when they start the process of cell division they generate a new cell that preserves the characteristics of the original and remains "dormant" until receiving a new stimulus that induces it to multiply and give rise to the same or even different cell lines; (2) Multipotentiality, ability of a cell to differentiate into different mature cells distinct from their tissue of origin such as adipocytes, chondrocytes, osteocytes, myoblasts, cardiomyocytes, neurons, astrocytes, myocytes, tenocytes, etc; (3) Functional viability, ability to perform the physiological functions of the cell type to which they belong.

Mesenchymal cells can be obtained from various adult tissue such as bone marrow, placenta<sup>[67]</sup>, umbilical cord [68], skeletal muscle<sup>[75]</sup>, synovial capsule<sup>[76]</sup>, synovial fluid<sup>[77]</sup>, and adipose tissue among others<sup>[78,79]</sup>. Mesenchymal cells from adipose tissue have a number of advantages over other cell types because adipose tissue is abundant and easy to obtain, can be obtained in large amounts, using local anesthesia and causing minimal discomfort<sup>[80]</sup>. Moreover, these cells have a high capacity for proliferation in vitro, have a fibroblastic morphology and adhere well to the culture plate. They express a specific set of known markers, and have a low risk of rejection<sup>[81]</sup>.

For ease of preparation, low risk of creating tumors, absence of rejection by the individual, their immunosuppressive and anti-inflammatory properties and that they do not pose ethical problems make them very good candidates for the treatment of different pathologies<sup>[82]</sup>. In human medicine, cell therapy is increasingly used for conditions such as hematologic malignancies and correction

of hereditary disorders in cells derived from bone marrow such as leukemias, myelomas, lymphomas, aplastic anemia, myelodysplastic syndromes and various genetic diseases among others<sup>[83]</sup>. They are also used in cardiovascular regenerative medicine, which could increase myocardial function after myocardial infarction<sup>[84]</sup>, in inflammatory bowel disease, where there are preliminary data on their ability to repair ulcers and fistulas<sup>[85]</sup>, diabetes mellitus, where they could improve glycemic profile by an increase in the number of pancreatic<sup>[86]</sup>, islets in musculoskeletal diseases such as osteopetrosis, osteogenesis imperfecta, infantile hypophosphatasia, arthritis, etc.<sup>[87-89]</sup>, in ocular pathologies such as trauma, burns, chemicals, Stevens Johnson syndrome or ocular pemphigoid and neurological diseases such as Parkinson's disease, amyotrophic lateral sclerosis, Alzheimer's disease, multiple sclerosis, stroke or spinal cord injury<sup>[90]</sup>.

In veterinary medicine there are studies with favorable results in dogs with osteoarthritis<sup>[58,59,78,91]</sup>, dogs with osteonecrosis of femoral head<sup>[92]</sup>, rats with neurological problems<sup>[93]</sup>, tendinitis and arthritic conditions in horses<sup>[61]</sup> and goats<sup>[94]</sup>.

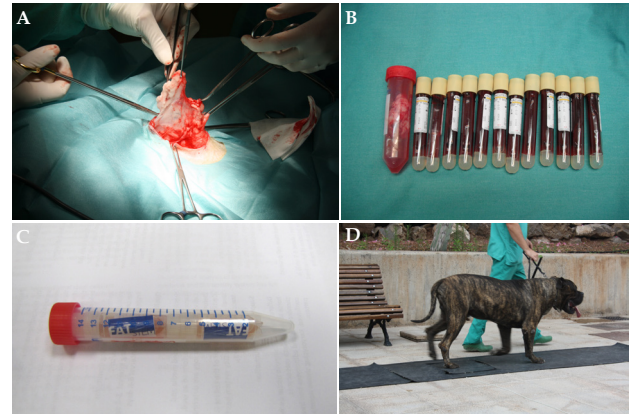
## MESENCHYMAL CELLS IN OSTEOARTHRITIS

As mentioned above, degenerative joint disease is one of the most prevalent diseases today in both humans and animals, which is why cell therapy is increasingly widespread in the field of traumatology and orthopedics for the treatment of pathologies such as osteoarthritis<sup>[7]</sup>.

Initially, it was believed that adult stem cells could only differentiate into the cell lineage to which they belonged, but today it has been shown that with different specific signals, these cells can be programmed to give rise to other organ specific cells with a different phenotype to the precursor. For this to happen it must overcome a number of barriers that exist in organisms *in vivo*, such as easy accessibility to sufficient cell concentration at the site of repair, and the generation of signals specific to the area to be treated to adequately direct the cells<sup>[70]</sup>. This statement opens a new insight into the use of adult stem cells because they can be obtained from various tissues with a simple and minimally invasive technique resulting in minimal harm to the patient without risk of rejection due to its autologous nature and also possess low immunogenicity, making them ideal candidates as a treatment of osteoarthritis<sup>[78,95]</sup>.

The treatment of degenerative joint disease with mesenchymal cells has different effects, on the one hand reduces pain and dependency on drugs, plus cells secrete proteins that are known for their anti-inflammatory potential, they also have several minimizer proteins of various stress markers in the surrounding cells, and finally the cells are installed in the surrounding tissues and generate new cartilage, thus slowing the degeneration and promoting intra-articular healing, thereby improving the general condition of the patients<sup>[91]</sup>.

There are various studies in different animal species that demonstrate the efficacy of this therapy in the treatment of degenerative articular cartilage lesions like the study performed by Mokbel *et al.*, who used mesenchymal cells in the treatment of osteoarthritis in donkeys demonstrating a significant improvement in the clinical signs as well as radiological exams, along with histopathological changes observing mesenchymal cells integrated in the cartilage surface and interior maintaining as mesenchymal cells or differentiating into chondrocytes<sup>[96]</sup>. Other studies were performed in rats with knee cartilage lesions treated with intra-articular injection of mesenchymal cells, showing that these cells are mobilized to the affected tissues and contribute to tissue regeneration<sup>[97,98]</sup>. Lee and colleagues used autologous mesenchymal cells in porcine knees



**Figure 3** A: Extraction of inguinal adipose tissue for cell culture. B: Sample Shipping Kit for obtaining mesenchymal cells from adipose tissue.

to regenerate experimentally created defects in the cartilage tissue, obtaining enhanced repair in the experimental group compared to the control group<sup>[99]</sup>. In domestic animals it has also been shown to be effective for use in this pathology, and Black *et al.* have conducted various studies in dogs with osteoarthritis in hip and knee, which after being treated intra-articularly with adipose derived mesenchymal stem cells observed a reduction of pain in the patients, as well as an improvement in lameness and increased range of motion<sup>[78,91]</sup>. In our research at the García Cugat Foundations' Surgery and Regenerative Medicine department at the CEU Cardinal Herrera University several studies in dogs with degenerative joint disease of the hip, elbow and knee are being carried out with intra-articular injections of inguinal adipose derived mesenchymal stem cells. Preliminary results at 6 months show improved clinical signs, as well as an increased range of joint mobility and decreased pain, resulting in a high satisfaction to the treatment by owners<sup>[58,59]</sup> (Figure 3). In some patients, this improvement has been quantified objectively using a force platform analysis obtaining favorable results in the evaluated parameters<sup>[100]</sup>.

Moreover, there are studies where this cell migration to the cartilage defect and its repair are not so obvious, like the study by Murphy and colleagues where, after injecting mesenchymal cells in knees affected by OA in goats, which was induced by meniscal tear and resection of the anterior cruciate ligament, did not observe mobilization of these cells into cartilage, but rather to the meniscus. Thus, at 6 weeks meniscus repair was observed but not in the articular cartilage or ligament, although this treatment did appear to stop the progressive destruction of articular cartilage<sup>[94]</sup>.

In human several studies support the effectiveness of this treatment both with mesenchymal cells and hematopoietic cells. In a study conducted on 24 patients with osteoarthritis of the knee infiltrated with bone marrow derived mesenchymal stem cells histological and arthroscopic improvement was observed<sup>[101]</sup>, or the study by Kudora *et al.* who concluded that transplantation of autologous bone marrow cells promotes repair of large focal articular cartilage defects in young and active<sup>[102]</sup> patients. Studies in human medicine in patients with osteoarthritis treated with mesenchymal cells also obtained good results, reducing symptoms and thereby increasing the satisfaction to the treatment, further demonstrating the regeneration potential induced in cartilage<sup>[103-105]</sup>.

Considering these studies, one can say that adult stem cells have the potential to differentiate into multiple lineages and integrate into joints affected by osteoarthritis. These cells are found in variety of tissues and are able to migrate to sites of injury where they exert a

substantial immunomodulatory and anti-inflammatory effect that is accomplished through direct cell interaction or via soluble mediators. The use of mesenchymal stem cells in tissue regeneration represents a promising avenue for the treatment of degenerative cartilage diseases opening a wide field of study on its role in the pathophysiology of osteoarthritis<sup>[106]</sup>.

With the aforementioned one can conclude that both PRP and MSCs, are a novel treatment that generate high hopes to overcoming the regeneration of articular cartilage in OA, while maintaining or improving the structure and joint function without the need for more aggressive techniques and cause further damage to the body. Notably, both PRGF therapy and aMSCs improved the pathology, that is, on one hand clinical symptoms improve, and furthermore, achieve an improvement on the structural damage caused by the disease<sup>[103,107]</sup>.

As for the advantages and disadvantages of each technique, it is important to note that PRP has the drawback of a shorter duration of effect, but has the important advantage of the lower cost method and a less invasive technique and therefore easier for outpatient use. In this case PRP treatment can be optimized with serial injections over time to obtain better results<sup>[107]</sup>. In the case of mesenchymal cells, their main drawback is the need to perform minor surgery under general anesthesia to obtain the tissue, with the advantages being the therapeutic benefit achieved. We believe that a possibility of optimizing this treatment is to create a bank of autologous cells, so they are available without any further surgeries. This option would bring the treatment to the outpatient clinic<sup>[108]</sup>.

## CONFLICT OF INTERESTS

There are no conflicts of interest with regard to the present study.

## REFERENCES

- Hunter DJ. Osteoarthritis. *Best Pract Res Clin Rheumatol* 2011; **25(6)**: 801-814.
- Rychel JK. Diagnosis and treatment of osteoarthritis. *Top Companion Anim Med* 2010; **25(1)**: 20-25
- Malek S, Sample SJ, Schwartz Z, Nemke B, Jacobson PB, Cozzi EM, Schaefer SL, Bleedorn JA, Holzman G, Muir P. Effect of analgesic therapy on clinical outcome measures in a randomized controlled trial using client-owned dogs with hip osteoarthritis. *BMC Vet Res* 2012; **8**: 185
- Juni P, Reichenbach S, Dieppe P. Osteoarthritis: rational approach to treating the individual. *Best Pract Res Clin Rheumatol* 2006; **20(4)**: 721-740
- van der Kraan PM. Osteoarthritis year 2012 in review: biology. *Osteoarthritis Cartilage* 2012; **20(12)**: 1447-1450
- Zanasi S, Brittberg M, Marcacci M. Basic Science, Clinical Repair and Reconstruction of Articular Cartilage Defects: Current Status and Prospects. First Edition ed. Vol. Vol.I. 2006, Bologna, Italy: Timeo Editore- Bologna. 600.
- Diekman BO, Guilak F. Stem cell-based therapies for osteoarthritis: challenges and opportunities. *Curr Opin Rheumatol* 2013; **25(1)**: 119-126
- Buckwalter JA, Mankin HJ, Grodzinsky AJ. Articular cartilage and osteoarthritis. *Instr Course Lect* 2005; **54**: 465-480
- Mortellaro CM. Pathophysiology of osteoarthritis. *Vet Res Commun* 2003; **27 Suppl 1**: 75-78.
- Brew K, Dinakarandian D, Nagase H. Tissue inhibitors of metalloproteinases: evolution, structure and function. *Biochim Biophys Acta* 2000; **1477(1-2)**: 267-283
- Goldring MB. Chondrogenesis, chondrocyte differentiation, and articular cartilage metabolism in health and osteoarthritis. *Ther Adv Musculoskelet Dis* 2012; **4(4)**: 269-285
- Goldring MB, Otero M, Plumb DA, Dragomir C, Favero M, El Hachem K, Hashimoto K, Roach HI, Olivotto E, Borzi RM, Marcu KB. Roles of inflammatory and anabolic cytokines in cartilage metabolism: signals and multiple effectors converge upon MMP-13 regulation in osteoarthritis. *Eur Cell Mater* 2011; **21**: 202-220
- Goldring MB, Marcu KB. Cartilage homeostasis in health and rheumatic diseases. *Arthritis Res Ther* 2009; **11(3)**: 224
- Leong DJ, Hardin JA, Cobelli NJ, Sun HB. Mechanotransduction and cartilage integrity. *Ann N Y Acad Sci* 2011; **1240**: 32-37
- Leong DJ, Li YH, Gu XI, Sun L, Zhou Z, Nasser P, Laudier DM, Iqbal J, Majeska RJ, Schaffler MB, Goldring MB, Cardoso L, Zaidi M, Sun HB. Physiological loading of joints prevents cartilage degradation through CITED2. *FASEB J* 2011; **25(1)**: 182-191
- Rialland P, Bichot S, Moreau M, Guillot M, Lussier B, Gauvin D, Martel-Pelletier J, Pelletier JP, Troncy E. Clinical validity of outcome pain measures in naturally occurring canine osteoarthritis. *BMC Vet Res* 2012; **8**: 162
- Bennett D, Zainal Ariffin SM, Johnston P. Osteoarthritis in the cat: 1. how common is it and how easy to recognise? *J Feline Med Surg* 2012; **14(1)**: 65-75
- Todhunter R, Johnston S. Osteoarthritis, in *Tratado de Cirugía en Pequeños Animales*, D. Slatter, Editor 2006, Inter-Médica: Buenos Aires, República Argentina. p. 2515-2561
- Reid MC, Shengelia R, Parker SJ. Pharmacologic Management of Osteoarthritis-Related Pain in Older Adults: A Review Shows that Many Drug Therapies Provide Small-to-Modest Pain Relief. *HSS J* 2012; **8(2)**: 159-164
- Godley DR. Assessment, diagnosis, and treatment of developmental dysplasia of the hip. *JAAPA* 2013; **26(3)**: 54-58
- Merashly M, Uthman I. Management of knee osteoarthritis: an evidence-based review of treatment options. *J Med Liban* 2012; **60(4)**: 237-242
- Allen MJ. Advances in total joint replacement in small animals. *J Small Anim Pract* 2012; **53(9)**: 495-506
- Nganvongpanit K, Itthiarbha A, Ong-Chai S, Kongtawelert P. Evaluation of serum chondroitin sulfate and hyaluronan: biomarkers for osteoarthritis in canine hip dysplasia. *J Vet Sci* 2008; **9(3)**: 317-325
- Cheng OT, Souzdanitski D, Vrooman B, Cheng J. Evidence-based knee injections for the management of arthritis. *Pain Med* 2012; **13(6)**: 740-753
- Hameed F, Ihm J. Injectable medications for osteoarthritis. *PM R* 2012; **4(5 Suppl)**: S75-81
- Giannini S, Buda R, Cavallo M, Ruffilli A, Cenacchi A, Cavallo C, Vannini F. Cartilage repair evolution in post-traumatic osteochondral lesions of the talus: from open field autologous chondrocyte to bone-marrow-derived cells transplantation. *Injury* 2010; **41(11)**: 1196-1203
- Ahmad Z, Howard D, Brooks RA, Wardale J, Henson FM, Getgood A, Rushton N. The role of platelet rich plasma in musculoskeletal science. *JRSM Short Rep* 2012; **3(6)**: 40
- Anitua E. Plasma rich in growth factors: preliminary results of use in the preparation of future sites for implants. *Int J Oral Maxillofac Implants* 1999; **14(4)**: 529-535
- Marx RE, Carlson ER, Eichstaedt RM, Schimmele SR, Strauss JE, Georgeff KR. Platelet-rich plasma: Growth factor enhancement for bone grafts. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 1998; **85(6)**: 638-646
- Grageda E. Platelet-rich plasma and bone graft materials: a review and a standardized research protocol. *Implant Dent* 2004; **13(4)**: 301-309



- 31 Canalis E. Clinical review 35: Growth factors and their potential clinical value. *J Clin Endocrinol Metab* 1992; **75**(1): 1-4
- 32 Reed, G.L., M.L. Fitzgerald, and J. Polgar, Molecular mechanisms of platelet exocytosis: insights into the “secrete” life of thrombocytes. *Blood* 2000; **96**(10): 3334-3342
- 33 Anitua E. The use of plasma-rich growth factors (PRGF) in oral surgery. *Pract Proced Aesthet Dent* 2001; **13**(6): 87-93; quiz 487-493
- 34 Ouyang XY, Qiao J. Effect of platelet-rich plasma in the treatment of periodontal intrabony defects in humans. *Chin Med J (Engl)* 2006; **119**(18): 1511-1521
- 35 Canalis E, McCarthy T, Centrella M. Growth factors and the regulation of bone remodeling. *J Clin Invest* 1988; **81**(2): 277-281
- 36 Civinini, R., Nistri L, Martini C, Redl B, Ristori G, Innocenti M. Growth factors in the treatment of early osteoarthritis. *Clin Cases Miner Bone Metab* 2013; **10**(1): 26-29
- 37 Civinini R, Macera A, Nistri L, Redl B, Innocenti M. The use of autologous blood-derived growth factors in bone regeneration. *Clin Cases Miner Bone Metab* 2011; **8**(1): 25-31
- 38 Ferrari M, Zia S, Valbonesi M, Henriquet F, Venere G, Spagnolo S, Grasso MA, Panzani I. A new technique for hemodilution, preparation of autologous platelet-rich plasma and intraoperative blood salvage in cardiac surgery. *Int J Artif Organs* 1987; **10**(1): 47-50
- 39 Whitman DH, Berry RL, Green DM. Platelet gel: an autologous alternative to fibrin glue with applications in oral and maxillofacial surgery. *J Oral Maxillofac Surg* 1997; **55**(11): 1294-1299
- 40 Floryan KM, Berghoff WJ. Intraoperative use of autologous platelet-rich and platelet-poor plasma for orthopedic surgery patients. *AORN J* 2004; **80**(4): 668-74, quiz 675-8
- 41 Sampson S, Gerhardt M, Mandelbaum B. Platelet rich plasma injection grafts for musculoskeletal injuries: a review. *Curr Rev Musculoskelet Med* 2008; **1**(3-4): 165-174
- 42 Marx, R.E., Platelet-rich plasma (PRP): what is PRP and what is not PRP? *Implant Dent* 2001; **10**(4): 225-228
- 43 Anitua E, Sánchez M, Orive G, Andia I. Delivering growth factors for therapeutics. *Trends Pharmacol Sci* 2008; **29**(1): 37-41
- 44 Anitua, E., Sánchez M, Orive G, Andia I. The potential impact of the preparation rich in growth factors (PRGF) in different medical fields. *Biomaterials* 2007; **28**(31): 4551-4560
- 45 Hunter DJ, Felson DT., Osteoarthritis. *BMJ* 2006; **332**(7542): 639-642
- 46 Fortier LA, Barker JU, Strauss EJ, McCarrel TM, Cole BJ. The role of growth factors in cartilage repair. *Clin Orthop Relat Res* 2011; **469**(10): 2706-2715
- 47 Goldring, M.B., K. Tsuchimochi, and K. Ijiri, The control of chondrogenesis. *J Cell Biochem* 2006; **97**(1): 33-44
- 48 Shi S, Mercer S, Eckert GJ, Trippel SB. Growth factor regulation of growth factors in articular chondrocytes. *J Biol Chem* 2009; **284**(11): 6697-6704
- 49 Anitua E, Sánchez M, Orive G, Padilla S. A biological therapy to osteoarthritis treatment using platelet-rich plasma. *Expert Opin Biol Ther* 2013; **13**(8): 1161-1172
- 50 Anitua E, Andia I, Ardanza B, Nurden P, Nurden AT. Autologous platelets as a source of proteins for healing and tissue regeneration. *Thromb Haemost* 2004; **91**(1): 4-15
- 51 Anitua E, Prado R, Sanchez M, Orive G. Platelet-Rich Plasma: Preparation and Formulation. *Oper Tech Orthop* 2012; **22**: 25-32
- 52 Anitua E, Sanchez M, Orive G. Potential of endogenous regenerative technology for in situ regenerative medicine. *Adv Drug Deliv Rev* 2010; **62**(7-8): 741-752
- 53 Wang-Saegusa A, Cugat R, Ares O, Seijas R, Cuscó X, Garcia-Balletbó M. Infiltration of plasma rich in growth factors for osteoarthritis of the knee short-term effects on function and quality of life. *Arch Orthop Trauma Surg* 2011; **131**(3): 311-317
- 54 Sanchez M, Anitua E, Azofra J, Aguirre JJ, Andia I. Intra-articular injection of an autologous preparation rich in growth factors for the treatment of knee OA: a retrospective cohort study. *Clin Exp Rheumatol* 2008; **26**(5): 910-913
- 55 Kwon DR, Park GY, Lee SU. The effects of intra-articular platelet-rich plasma injection according to the severity of collagenase-induced knee osteoarthritis in a rabbit model. *Ann Rehabil Med* 2012; **36**(4): 458-465
- 56 Serra CI, Soler C, Carrillo JM, Sopena JJ, Redondo JI, Cugat R. Effect of autologous platelet-rich plasma on the repair of full-thickness articular defects in rabbits. *Knee Surg Sports Traumatol Arthrosc* 2012
- 57 Milano G, Sanna Passino E, Deriu L, Careddu G, Manunta L, Manunta A, Saccomanno MF, Fabbriani C. The effect of platelet rich plasma combined with microfractures on the treatment of chondral defects: an experimental study in a sheep model. *Osteoarthritis Cartilage* 2010; **18**(7): 971-780
- 58 Cuervo B. Effect of adipose mesenchymal stem cells and plasma rich in growth factors in the treatment of osteoarthritis in dogs. in Termis-eu 2013. 2013. Istanbul, Turkey
- 59 Cuervo B. Mesenchymal stem cells from adipose tissue and plasma rich in growth factors in degenerative joint disease in dogs. *Osteoarthritis Cartilage* 2013; **21** supplement: 119-120.
- 60 Silva RF, Carmona JU, Rezende CM. Intra-articular injections of autologous platelet concentrates in dogs with surgical reparation of cranial cruciate ligament rupture. A pilot study. *Vet Comp Orthop Traumatol* 2013; **26**(4): 285-290
- 61 Fortier LA, Travis AJ. Stem cells in veterinary medicine. *Stem Cell Res Ther* 2011; **2**(1): 9
- 62 Cahan P, Daley GQ. Origins and implications of pluripotent stem cell variability and heterogeneity. *Nat Rev Mol Cell Biol* 2013; **14**(6): 357-368
- 63 Yao S, Chen S, Clark J, Hao E, Beattie GM, Hayek A, Ding S. Long-term self-renewal and directed differentiation of human embryonic stem cells in chemically defined conditions. *Proc Natl Acad Sci U S A* 2006; **103**(18): 6907-6912
- 64 Hoffman LM, Carpenter MK. Characterization and culture of human embryonic stem cells. *Nat Biotechnol* 2005; **23**(6): 699-708
- 65 Aflatoonian B, Moore H. Human primordial germ cells and embryonic germ cells, and their use in cell therapy. *Curr Opin Biotechnol* 2005; **16**(5): 530-535
- 66 Zhu Y, Yang Y, Zhang Y, Hao G, Liu T, Wang L, Yang T, Wang Q, Zhang G, Wei J, Li Y. Placental mesenchymal stem cells of fetal and maternal origins demonstrate different therapeutic potentials. *Stem Cell Res Ther* 2014; **5**(2): 48
- 67 Timmins NE, Kiel M, Günther M, Heazlewood C, Doran MR, Brooke G, Atkinson K. Closed system isolation and scalable expansion of human placental mesenchymal stem cells. *Biotechnol Bioeng* 2012; **109**(7): 1817-1826
- 68 Yang S, Huang S, Feng C, Fu X. Umbilical cord-derived mesenchymal stem cells: strategies, challenges, and potential for cutaneous regeneration. *Front Med* 2012; **6**(1): 41-47
- 69 Verfaillie CM, Pera MF, Lansdorp PM. Stem cells: hype and reality. *Hematology Am Soc Hematol Educ Program* 2002; 369-391
- 70 Korbliing M, Estrov Z. Adult stem cells for tissue repair—a new therapeutic concept? *N Engl J Med* 2003; **349**(6): 570-582
- 71 Rosenthal N. Prometheus’s vulture and the stem-cell promise. *N Engl J Med* 2003; **349**(3): 267-274
- 72 Hanna JH, Saha K, Jaenisch R. Pluripotency and cellular reprogramming: facts, hypotheses, unresolved issues. *Cell* 2010; **143**(4): 508-525
- 73 Mizuno H, Tobita M, Uysal AC. Concise review: Adipose-derived stem cells as a novel tool for future regenerative medicine. *Stem Cells* 2012; **30**(5): 804-810

- 74 Kolios G, Moodley Y. Introduction to stem cells and regenerative medicine. *Respiration* 2013; **85(1)**: 3-10
- 75 Jackson WM, Lozito TP, Djouad F, Kuhn NZ, Nesti LJ, Tuan RS. Differentiation and regeneration potential of mesenchymal progenitor cells derived from traumatized muscle tissue. *J Cell Mol Med* 2011; **15(11)**: 2377-2388
- 76 Jones BA, Pei M. Synovium-derived stem cells: a tissue-specific stem cell for cartilage engineering and regeneration. *Tissue Eng Part B Rev* 2012; **18(4)**: 301-311
- 77 Jones EA, Crawford A, English A, Henshaw K, Mundy J, Corscadden D, Chapman T, Emery P, Hatton P, McGonagle D. Synovial fluid mesenchymal stem cells in health and early osteoarthritis: detection and functional evaluation at the single-cell level. *Arthritis Rheum* 2008; **58(6)**: 1731-1740
- 78 Black LL, Gaynor J, Adams C, Dhupa S, Sams AE, Taylor R, Harman S, Gingerich DA, Harman R. Effect of intraarticular injection of autologous adipose-derived mesenchymal stem and regenerative cells on clinical signs of chronic osteoarthritis of the elbow joint in dogs. *Vet Ther* 2008; **9(3)**: 192-200
- 79 Yarak S, Okamoto OK. Human adipose-derived stem cells: current challenges and clinical perspectives. *An Bras Dermatol* 2010; **85(5)**: 647-656
- 80 Noth U, Steinert AF, Tuan RS. Technology insight: adult mesenchymal stem cells for osteoarthritis therapy. *Nat Clin Pract Rheumatol* 2008; **4(7)**: 371-380
- 81 McIntosh KR, Frazier T, Rowan BG, Gimble JM. Evolution and future prospects of adipose-derived immunomodulatory cell therapeutics. *Expert Rev Clin Immunol* 2013; **9(2)**: 175-184
- 82 Lisianyi MI, [Mesenchymal stem cells and their immunological properties]. *Fiziol Zh* 2013; **59(3)**: 126-134
- 83 O'Brien JA, Rizzieri DA. Leukemic stem cells: a review. *Cancer Invest* 2013; **31(4)**: 215-220
- 84 Hare JM, Chaparro SV. Cardiac regeneration and stem cell therapy. *Curr Opin Organ Transplant* 2008; **13(5)**: 536-542
- 85 Garcia-Olmo, D., Garcia-Arranz M, Herreros D, Pascual I, Peiro C, Rodríguez-Montes JA. A phase I clinical trial of the treatment of Crohn's fistula by adipose mesenchymal stem cell transplantation. *Dis Colon Rectum* 2005; **48(7)**: 1416-1423
- 86 Lee, R.H., Seo MJ, Reger RL, Spees JL, Pulin AA, Olson SD, Prockop DJ. Multipotent stromal cells from human marrow home to and promote repair of pancreatic islets and renal glomeruli in diabetic NOD/scid mice. *Proc Natl Acad Sci U S A* 2006; **103(46)**: 17438-17443
- 87 Ma D, Mao t, [Cell-based approaches to promote bone regeneration in distraction osteogenesis]. *Zhongguo Xiu Fu Chong Jian Wai Ke Za Zhi* 2012; **26(12)**: 1512-1515
- 88 Singh JA. Stem cells and other innovative intra-articular therapies for osteoarthritis: what does the future hold? *BMC Med* 2012; **10**: 44
- 89 Chen FH, Tuan RS. Mesenchymal stem cells in arthritic diseases. *Arthritis Res Ther* 2008; **10(5)**: 223
- 90 Prosper F, Gavira JJ, Herreros J, Rábago G, Luquin R, Moreno J, Robles JE, Redondo P. [Cell transplant and regenerative therapy with stem cells]. *An Sist Sanit Navar* 2006; **29 Suppl 2**: 219-234
- 91 Black LL, Gaynor J, Gahring D, Adams C, Aron D, Harman S, Gingerich DA, Harman R. Effect of adipose-derived mesenchymal stem and regenerative cells on lameness in dogs with chronic osteoarthritis of the coxofemoral joints: a randomized, double-blinded, multicenter, controlled trial. *Vet Ther* 2007; **8(4)**: 272-284
- 92 Hang D, Wang Q, Guo C, Chen Z, Yan Z. Treatment of osteonecrosis of the femoral head with VEGF165 transgenic bone marrow mesenchymal stem cells in mongrel dogs. *Cells Tissues Organs* 2012; **195(6)**: 495-506
- 93 Liao CD, Zhang F, Guo RM, Zhong XM, Zhu J, Wen XH, Shen J. Peripheral nerve repair: monitoring by using gadofluorine M-enhanced MR imaging with chitosan nerve conduits with cultured mesenchymal stem cells in rat model of neurotmesis. *Radiology* 2012; **262(1)**: 161-171
- 94 Murphy JM, Fink DJ, Hunziker EB, Barry FP. Stem cell therapy in a caprine model of osteoarthritis. *Arthritis Rheum* 2003; **48(12)**: 3464-3474
- 95 Heng BC, Cao T, Lee EH. Directing stem cell differentiation into the chondrogenic lineage *in vitro*. *Stem Cells* 2004; **22(7)**: 1152-1167
- 96 Mokbel AN, El Tookhy OS, Shamaa AA, Rashed LA, Sabry D, El Sayed AM. Homing and reparative effect of intra-articular injection of autologous mesenchymal stem cells in osteoarthritic animal model. *BMC Musculoskelet Disord* 2011; **12**: 259
- 97 Agung M, Ochi M, Yanada S, Adachi N, Izuta Y, Yamasaki T, Toda K. Mobilization of bone marrow-derived mesenchymal stem cells into the injured tissues after intraarticular injection and their contribution to tissue regeneration. *Knee Surg Sports Traumatol Arthrosc* 2006; **14(12)**: 1307-1314
- 98 Nishimori M, Deie M, Kanaya A, Exham H, Adachi N, Ochi M. Repair of chronic osteochondral defects in the rat. A bone marrow-stimulating procedure enhanced by cultured allogenic bone marrow mesenchymal stromal cells. *J Bone Joint Surg Br* 2006; **88(9)**: 1236-1244
- 99 Lee KB, Hui JH, Song IC, Ardany L, Lee EH. Injectable mesenchymal stem cell therapy for large cartilage defects—a porcine model. *Stem Cells* 2007; **25(11)**: 2964-2971
- 100 Vilar JM, Morales M, Santana A, Spinella G, Rubio M, Cuervo B, Cugat R, Carrillo JM. Controlled, blinded force platform analysis of the effect of intraarticular injection of autologous adipose-derived mesenchymal stem cells associated to PRGF-Endoret in osteoarthritic dogs. *BMC Vet Res* 2013; **9**: 131
- 101 Wakitani S, Imoto K, Yamamoto T, Saito M, Murata N, Yoneda M. Human autologous culture expanded bone marrow mesenchymal cell transplantation for repair of cartilage defects in osteoarthritic knees. *Osteoarthritis Cartilage* 2002; **10(3)**: 199-206
- 102 Kuroda R, Ishida K, Matsumoto T, Akisue T, Fujioka H, Mizuno K, Ohgushi H, Wakitani S, Kurosaka M. Treatment of a full-thickness articular cartilage defect in the femoral condyle of an athlete with autologous bone-marrow stromal cells. *Osteoarthritis Cartilage* 2007; **15(2)**: 226-231
- 103 Emadedin M, Aghdami N, Taghiyar L, Fazeli R, Moghadasali R, Jahangir S, Farjad R, Baghaban Eslaminejad M. Intra-articular injection of autologous mesenchymal stem cells in six patients with knee osteoarthritis. *Arch Iran Med* 2012; **15(7)**: 422-428
- 104 Davatchi F, Abdollahi BS, Mohyeddin M, Shahram F, Nikbin B. Mesenchymal stem cell therapy for knee osteoarthritis. Preliminary report of four patients. *Int J Rheum Dis* 2011; **14(2)**: 211-215
- 105 Koh YG, Jo SB, Kwon OR, Suh DS, Lee SW, Park SH, Choi YJ. Mesenchymal stem cell injections improve symptoms of knee osteoarthritis. *Arthroscopy* 2013; **29(4)**: 748-755
- 106 MacFarlane RJ, Graham SM, Davies PS, Korres N, Tsouchnica H, Heliotis M, Mantalaris A, Tsiroidis E. Anti-inflammatory role and immunomodulation of mesenchymal stem cells in systemic joint diseases: potential for treatment. *Expert Opin Ther Targets* 2013; **17(3)**: 243-254
- 107 Andia I, Sanchez M, Maffulli N. Joint pathology and platelet-rich plasma therapies. *Expert Opin Biol Ther* 2012; **12(1)**: 7-22
- 108 Dhanasekaran M, Indumathi S, Poojitha R, Kanmani A, Rajkumar JS, Sudarsanam D. Plasticity and banking potential of cultured adipose tissue derived mesenchymal stem cells. *Cell Tissue Bank* 2012



Cuervo B *et al.* Treatment in osteoarthritis

**Peer reviewers:** Ray Marks, Professor, Director Center for Health Promotion, School of Health and Behavioral Sciences, Department of Health & Physical Education, Gerontological Services and Studies, York College, City University of New York, Jamaica, NY 11451, USA; Wing P. Chan, MD, Department of Radiology, Wan Fang Hospital, Taipei Medical University, 111

Hsing Long Road, Section 3, Taipei 116, Taiwan; Sheng-Ming Dai, Professor, Department of Rheumatology & Immunology, Changhai Hospital, Second Military Medical University, 168 Changhai Road, Shanghai 200433, China; Pauline Siew Mei Lai, Senior Lecturer, Department of Primary Care Medicine, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur, Malaysia.